Toxicity Studies with Ordnance-Contaminated Marine Sediments

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Presentation Outline

- Project history
- Ordnance marine toxicity database development
- Sediment survey and Toxicity Identification Evaluation (TIE) study
- Ordnance-spiked sediment toxicity studies
- Conclusions and recommendations for future studies with ordnance compounds

Project History

- Studies conducted during the early/mid 1990s had identified a number of ordnance compounds in sediments adjacent to several naval facilities in Puget Sound.

Project History

- Little or no marine toxicity data were available for these ordnance compounds.

- Washington state Dept. of Ecology had requested that these sediments be removed to below detectable levels (estimated cost ~$9M).

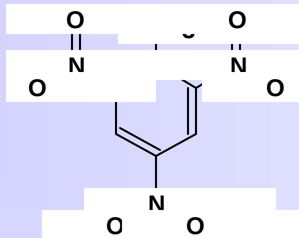
Ordnance Marine Toxicity Database Study Objectives

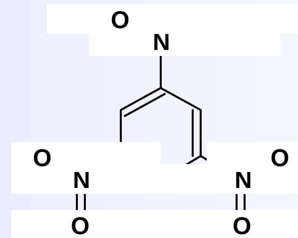
- Generate a database for toxicity of 8 ordnance compounds using 5 marine species and 9 toxicity test endpoints

- Identified the most sensitive species and endpoints and the relative toxicity of the different ordnance compounds

Ordnance Compounds of Concern

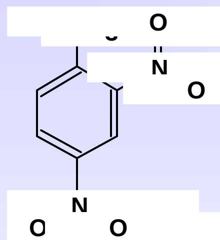
2, 4, 6-TNT

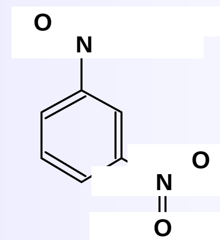




1, 3, 5-TNB

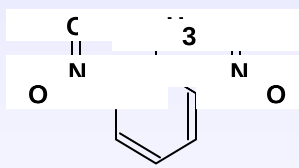
2, 4-DNT

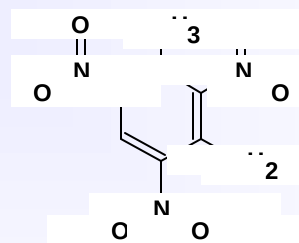




1, 3-DNB

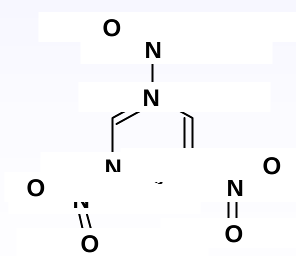
2, 6-DNT

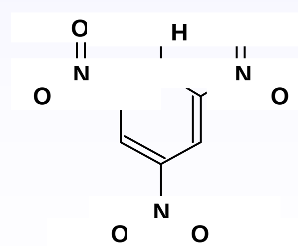




Tetryl

RDX





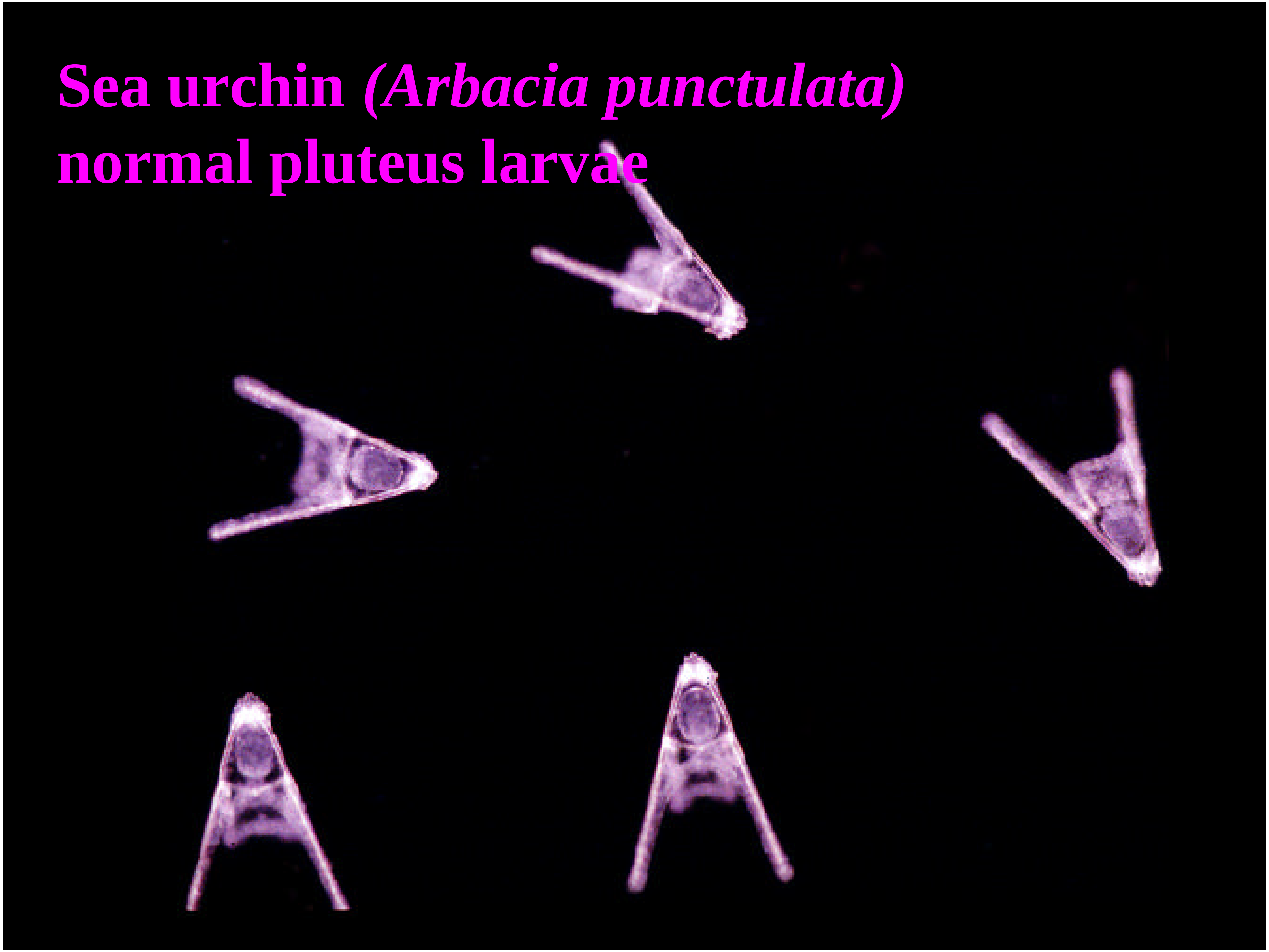
Picric Acid

Sea urchin *(Arbacia punctulata)* eggs

Not fertilized

Fertilized

Sea urchin *(Arbacia punctulata)* normal pluteus larvae



Polychaete (*Dinophilus gyrociliatus*)

Juvenile

Egg case with female and dwarf male eggs

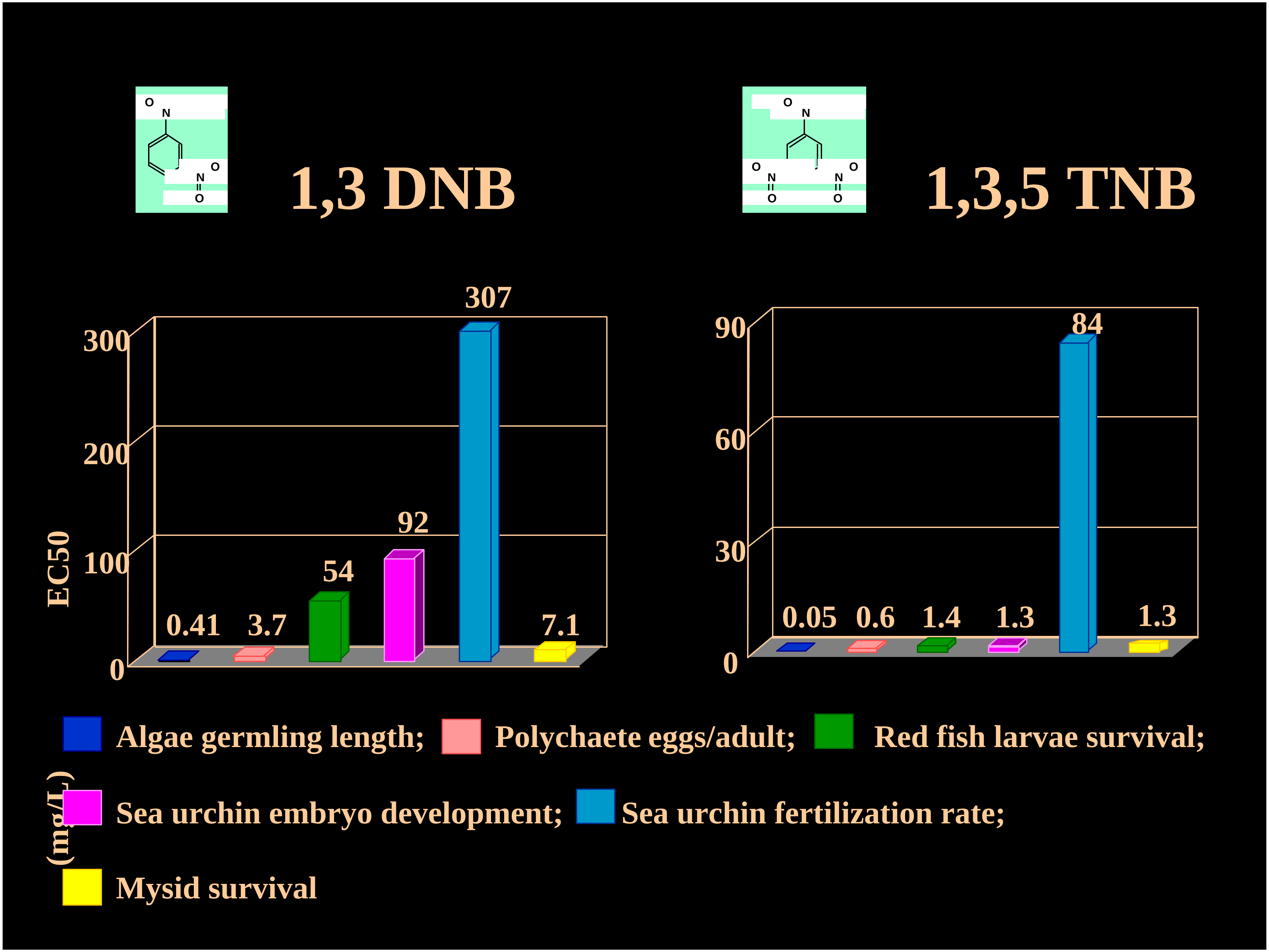
Opossum shrimp (mysid)

Red fish (*Sciaenops ocellatus*) normal and abnormal larvae, 24 hours after hatching

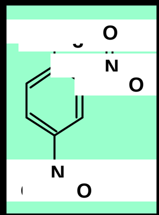
Macro-alga (*Ulva fasciata*)

One-cell germlings

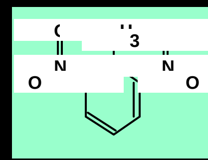
Multi-cell germlings



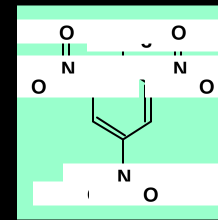
1,3 DNB
1,3,5 TNB
EC50 (mg/L)
300
200
100
0
0.41
3.7
54
92
307
7.1
90
60
30
0
0.05
0.6
1.4
1.3
84
1.3
Algae germling length;
Polychaete eggs/adult;
Red fish larvae survival;
Sea urchin embryo development;
Sea urchin fertilization rate;
Mysid survival



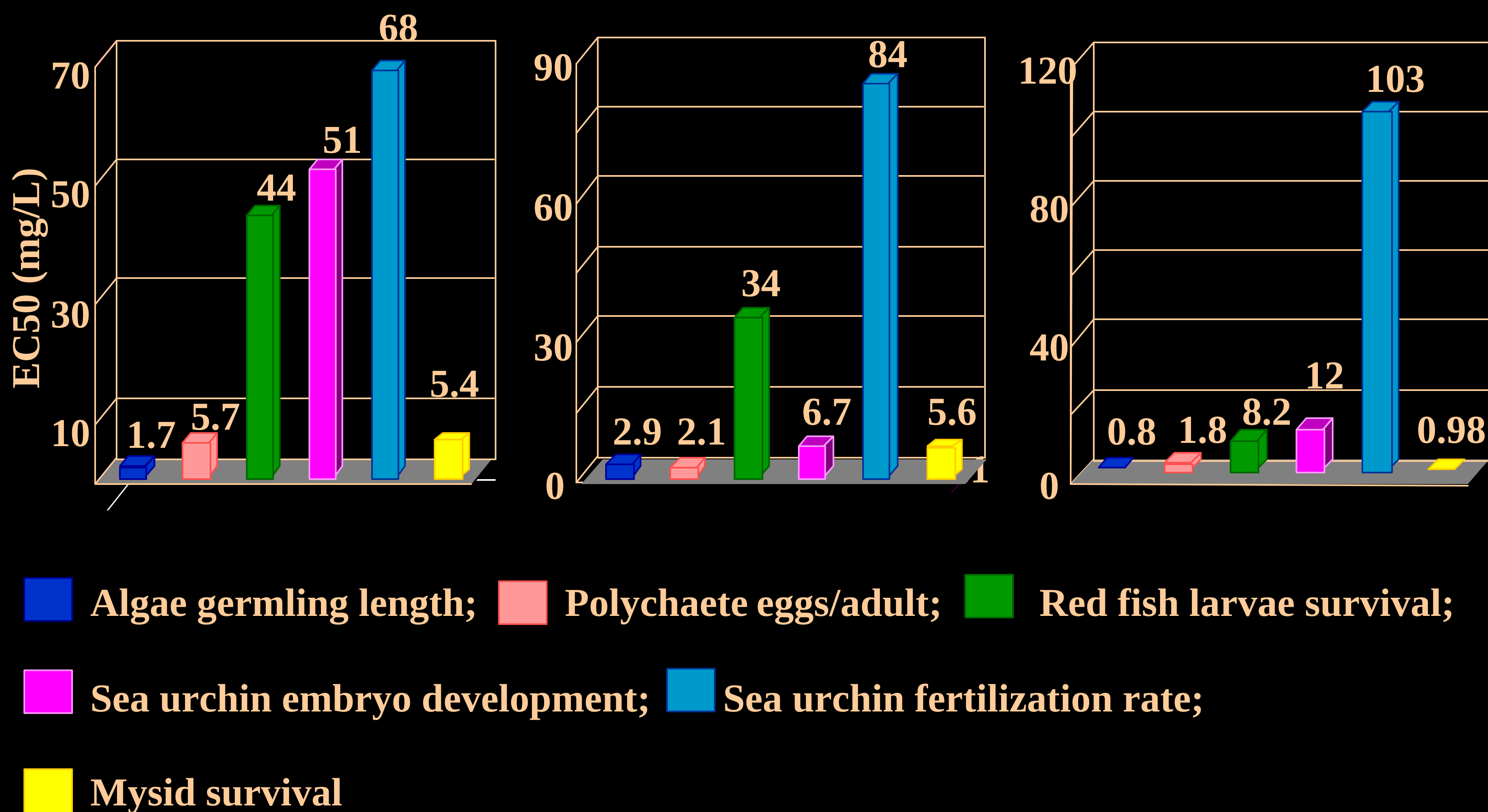
2,4-DNT

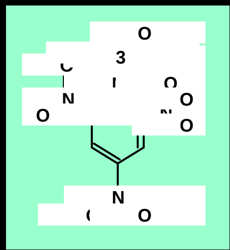


2,6-DNT

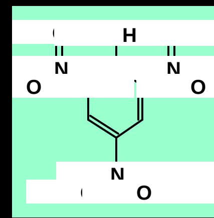


2,4,6-TNT

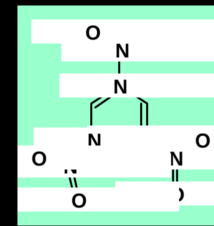




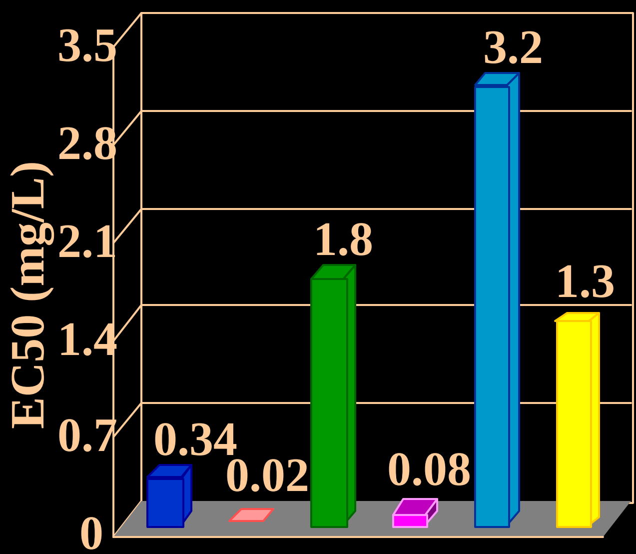
Tetryl

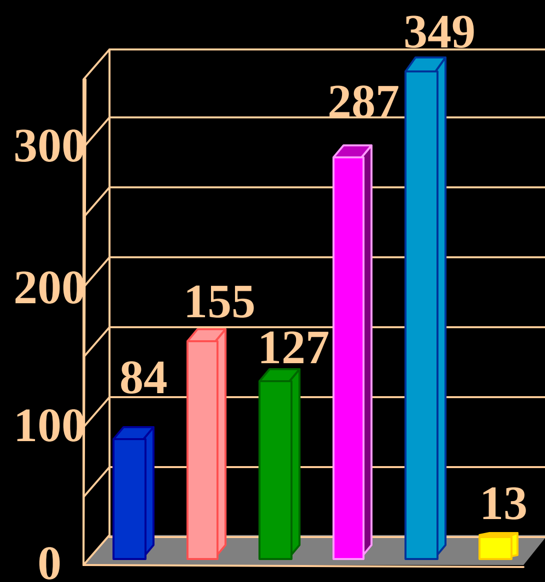


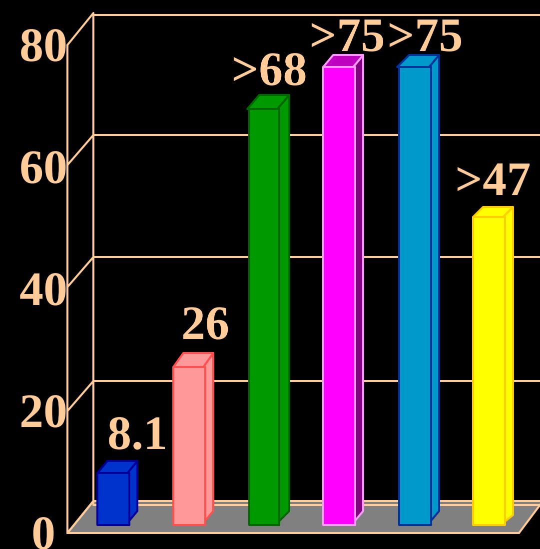
Picric Acid



RDX







Algae germling length; Polychaete eggs/adult; Red fish larvae survival;

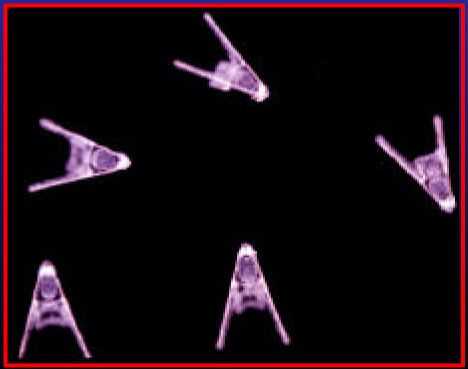
Sea urchin embryo development; Sea urchin fertilization rate;

Mysid survival

Most Sensitive Species and Endpoints

- Polychaete, *Dinophilus gyrociliatus*, reproduction

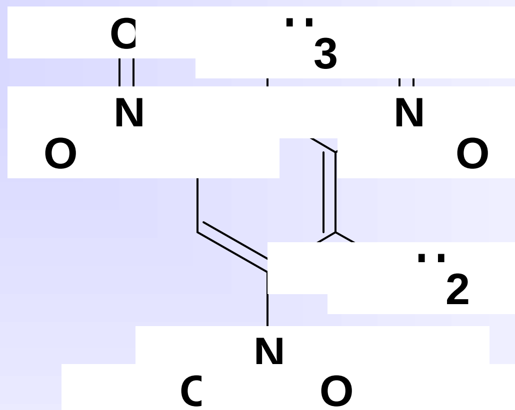
- Macro-alga, *Ulva fasciata*, germling growth



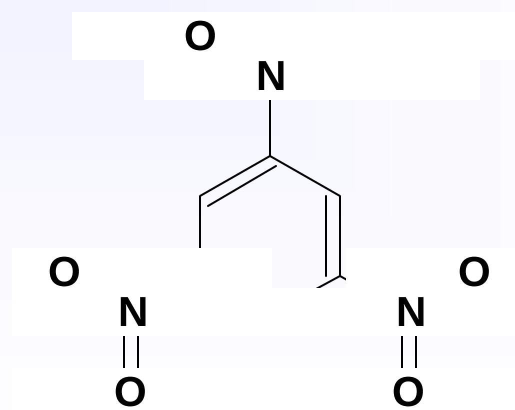
- Sea urchin, *Arbacia punctulata*, embryological development

Most Toxic Ordnance Compounds

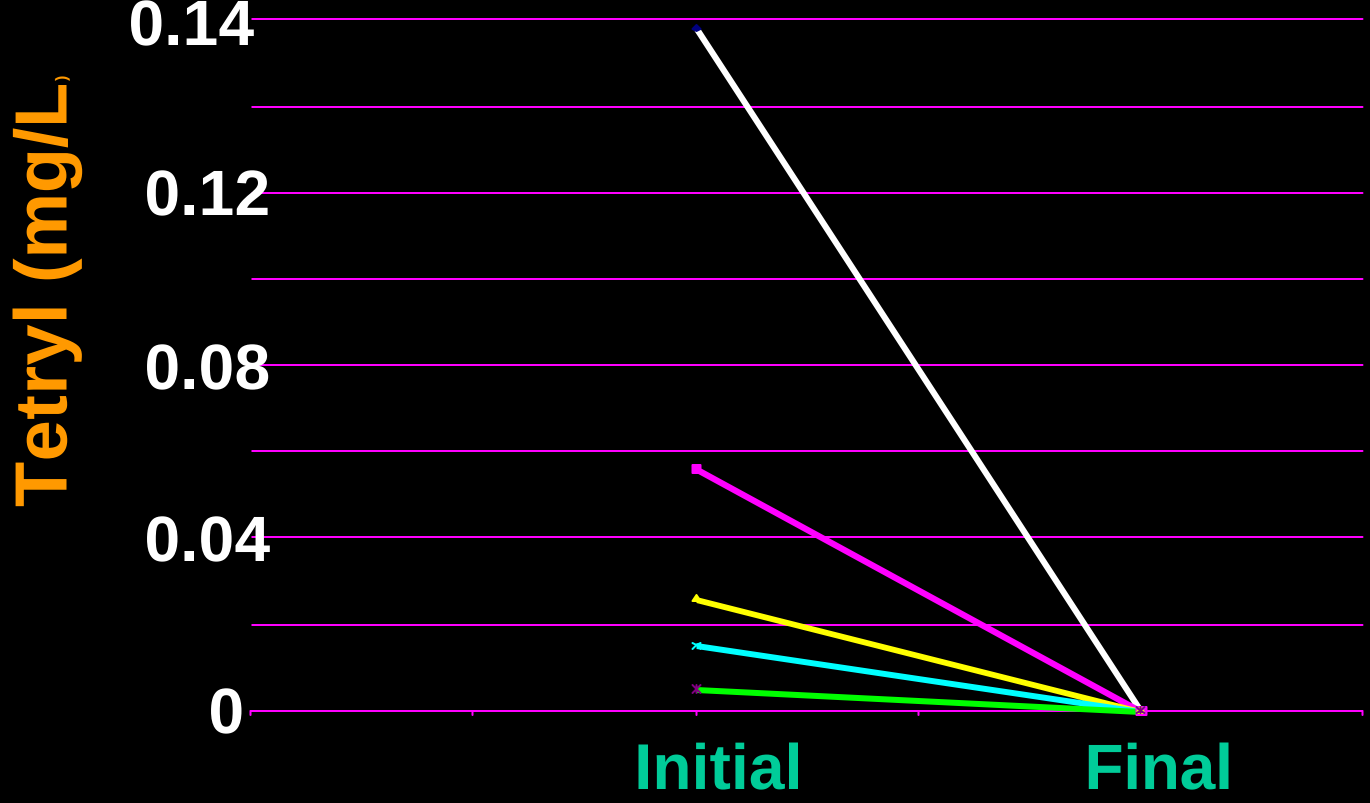
Tetryl



1,3,5-Trinitrobenzene

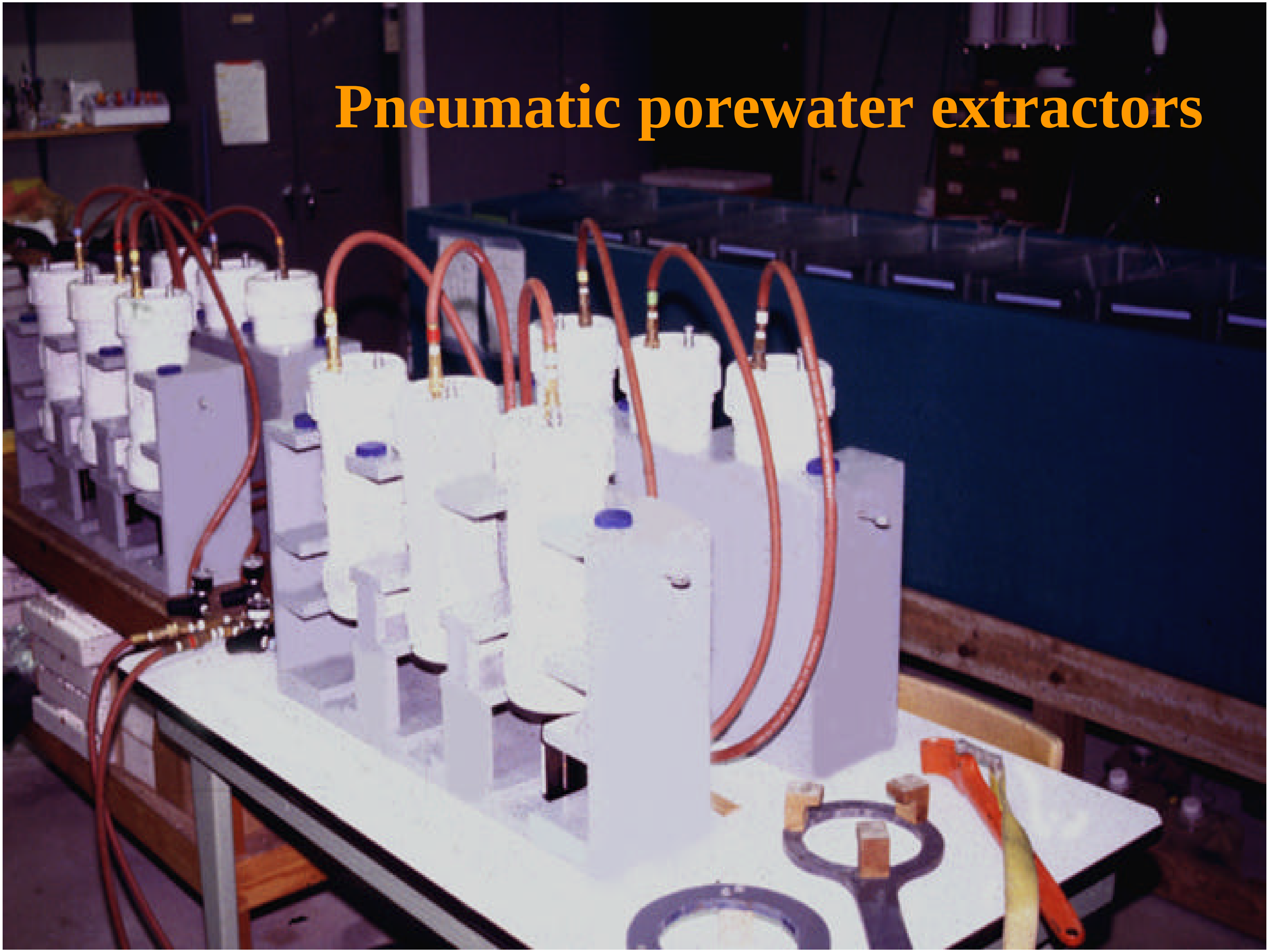


Polychaete test
Tetryl loss in 7 days



Objectives for Field Survey and TIE Study

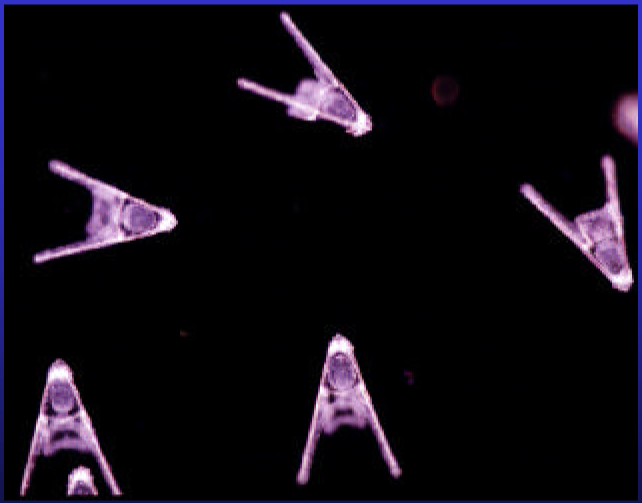
- Assess the degree and extent of toxicity in the vicinity of Jackson Park and Port Hadlock naval facilities using porewater toxicity tests
- Perform comprehensive chemical analyses on subset of sediments from survey based on toxicity test results
- Based on chemistry and toxicity data, select sites for TIE study


Pneumatic porewater extractors

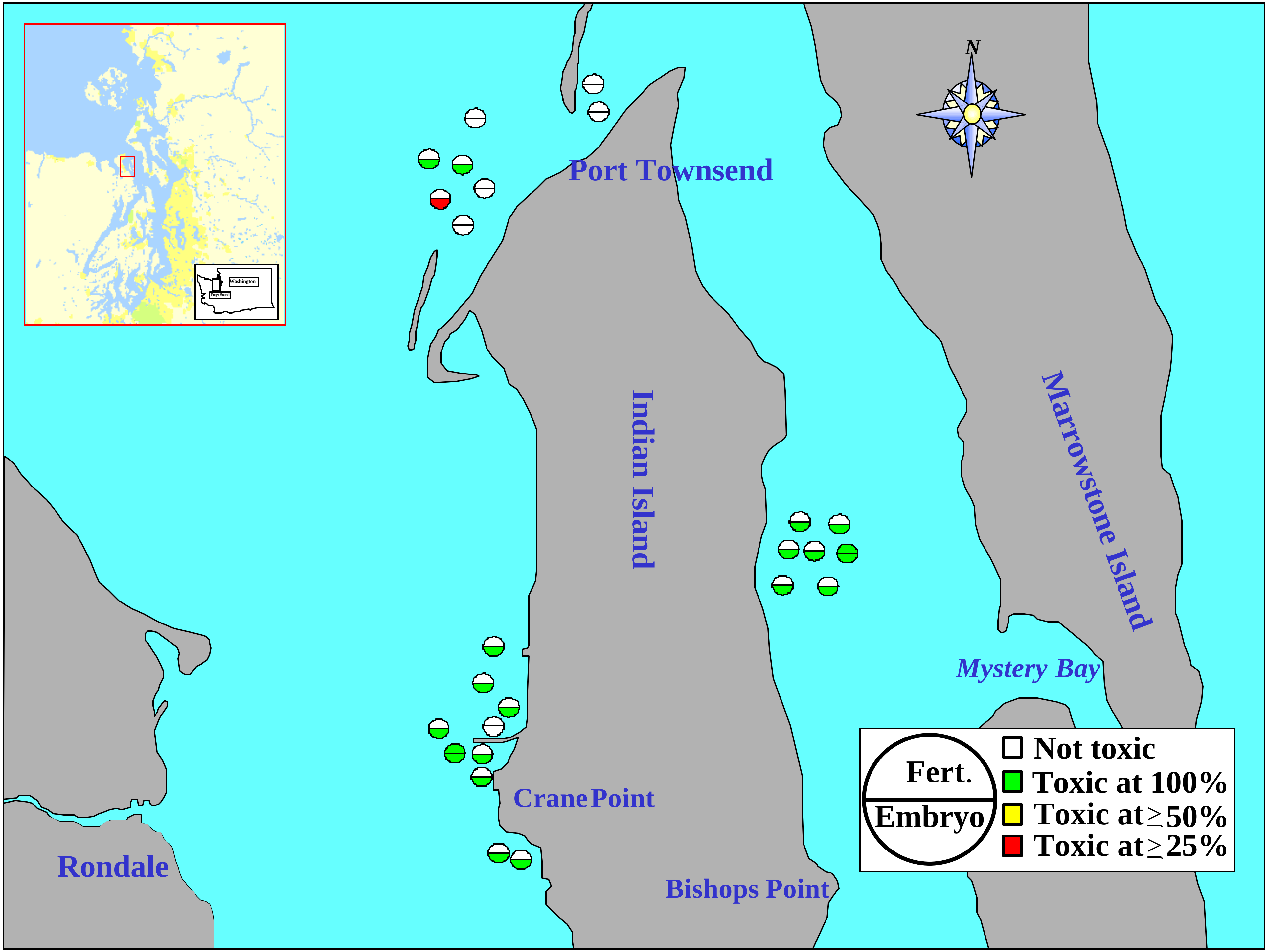
Inducing Sea Urchins to Spawn

Sea Urchin Test Setup

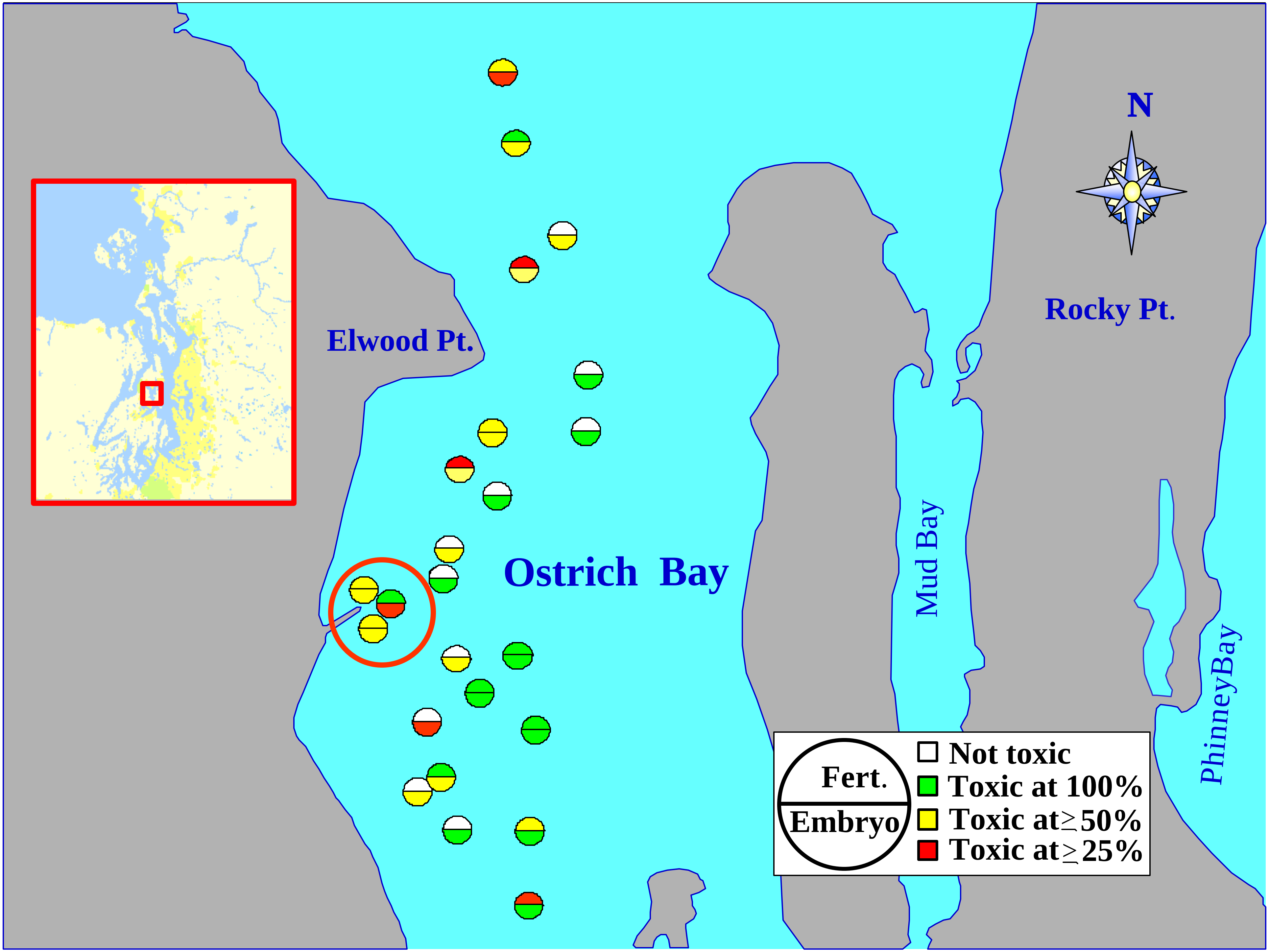
Unfertilized and Fertilized Egg



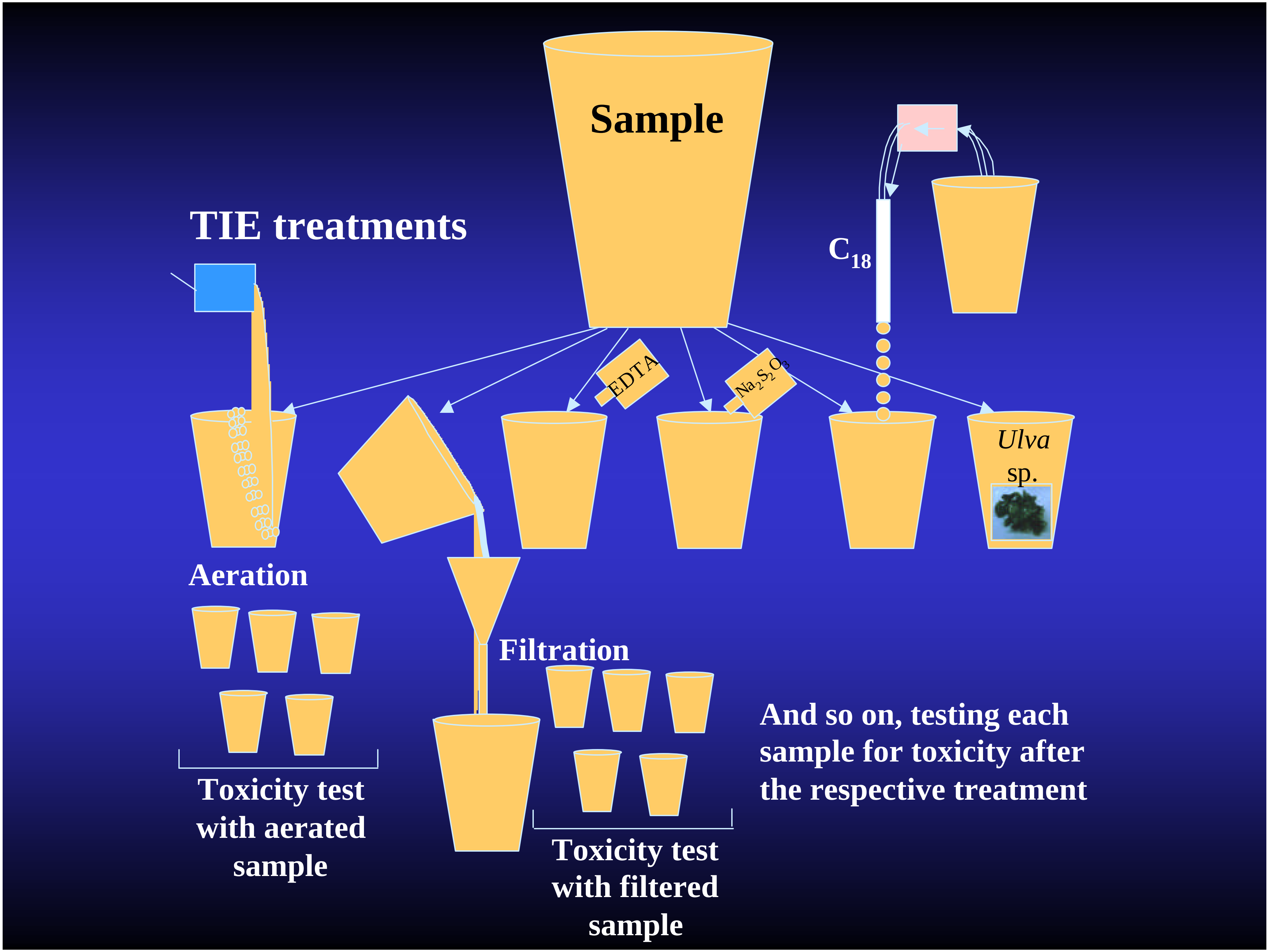
Arbacia Echinoplutei



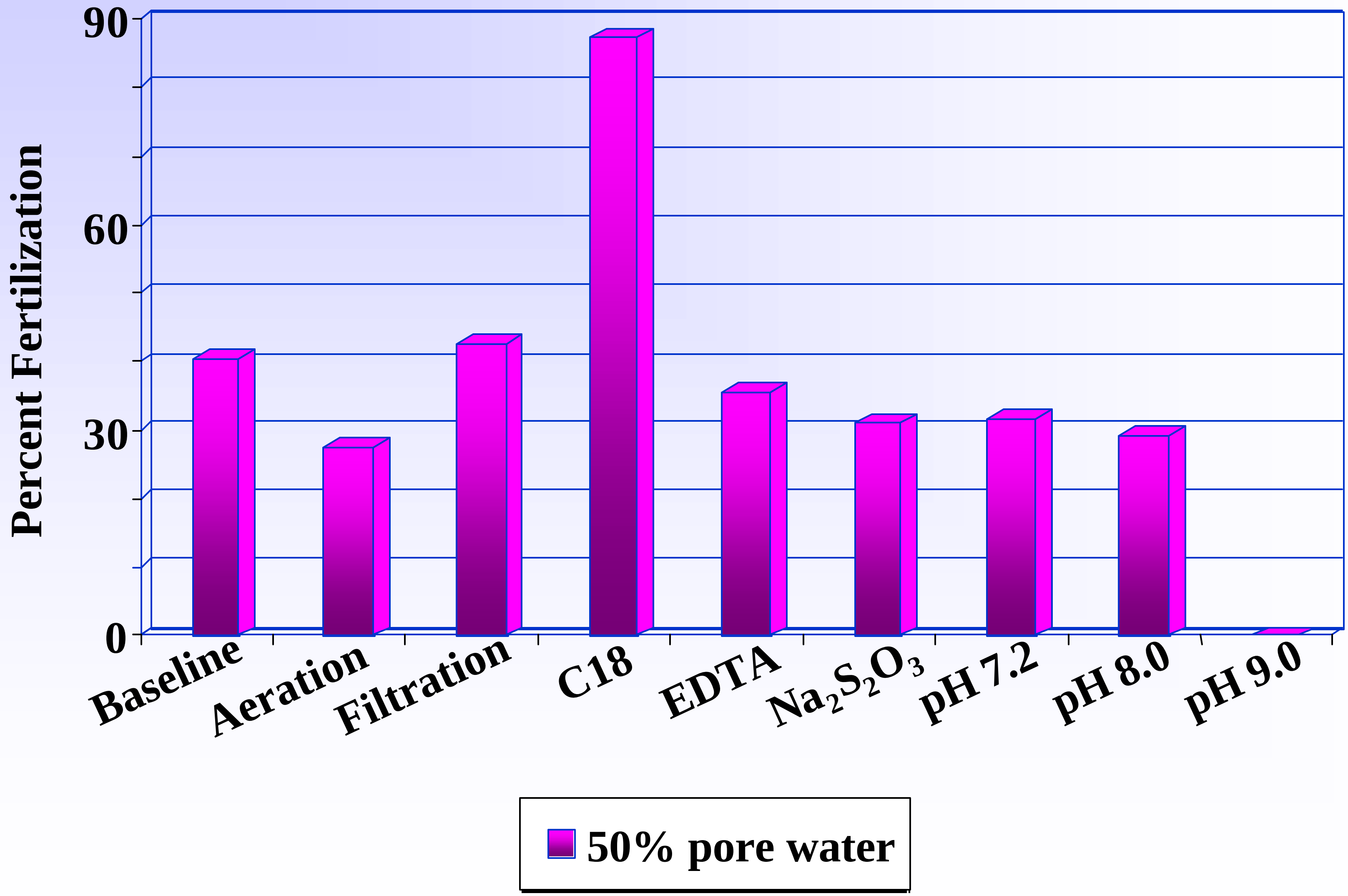
N
Port Townsend
Indian Island
Marrowstone Island
Mystery Bay
Crane Point
Bishops Point
Rondale
Washington
Puget Sound
Fert.
Embryo
Not toxic
Toxic at 100%
Toxic at ≥ 50%
Toxic at ≥ 25%



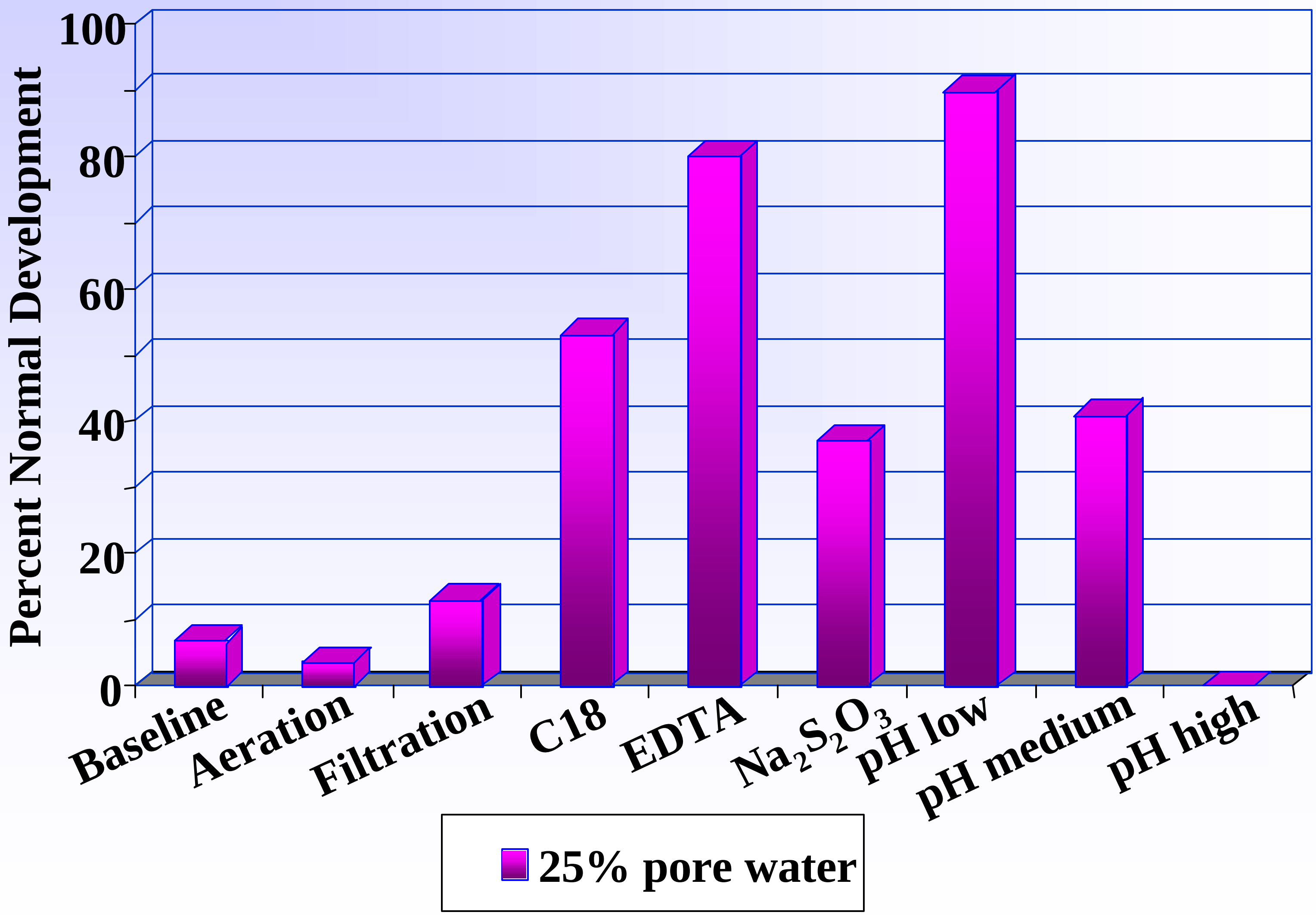
N
Rocky Pt.
Elwood Pt.
Ostrich Bay
Mud Bay
PhinneyBay
Fert.
Embryo
Not toxic
Toxic at 100%
Toxic at ≥50%
Toxic at ≥25%


Sample
TIE treatments
C18
EDTA
Na2S2O3
Ulva sp.
Aeration
Filtration
Toxicity test with aerated sample
Toxicity test with filtered sample
And so on, testing each sample for toxicity after the respective treatment

Toxicity Identification Evaluation (TIE): Ostrich Bay Sea Urchin Fertilization Results



Toxicity Identification Evaluation (TIE): Sea Urchin Embryological Development Results



Organics Analysis of Pore Water from Puget Sound TIE Site

Chemical	Fresh (:g/L)	Frozen (:g/L)
Phenol	1.5	1.5
Naphthalene	0.03	0.03
Phenanthrene	0.05	0.05
PCBs	ND	ND
Pesticides	ND	ND
14 Ordnance Compounds	ND	ND

Metals Analysis of Pore Water from Puget Sound TIE Site

Metal	Fresh (µg/L)	Frozen (µg/L)
As	8.0	4.3
Cd	0.08	0.04
Cu	0.3	0.2
Pb	0.14	0.06
Zn	1.1	0.6
Total BTs	0.120	0.118

Conclusions from Sediment Toxicity Survey and TIE Study

- Although porewater toxicity was observed at many of the sites studied, only very low levels of picric acid and 2,6-DNT were detected at several sites
- TIE studies indicate that ordnance compounds of concern were not responsible for observed toxicity which was due to unidentified organics, metals, and unionized ammonia.

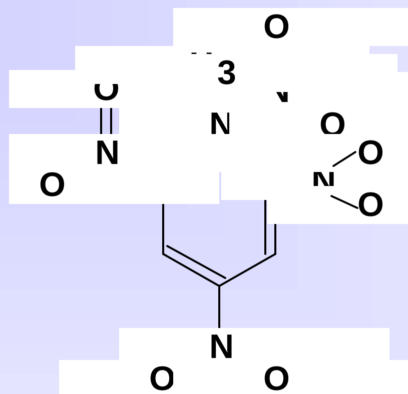
Objectives of Sediment Spiking Studies

- To identify the toxicity of ordnance compounds in spiked sediments and corresponding pore waters to marine organisms.

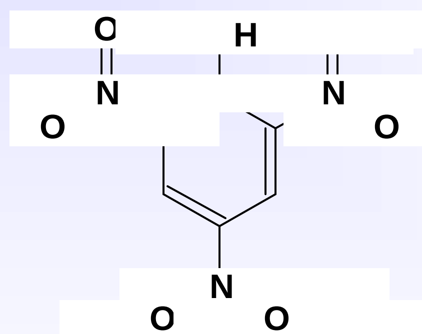
- To identify the role of different sediment features (grain size, TOC) on the toxicity and stability of ordnance compounds in spiked sediments and corresponding pore waters.

Ordnance compounds selected for sediment spiking studies

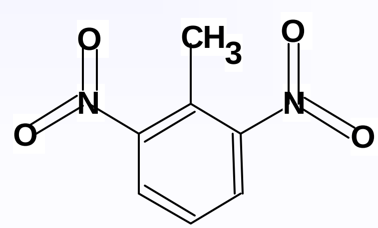
Tetryl



Picric acid



2,6-Dinitrotoluene



Two types of sediments selected for study

Carr Inlet, Puget Sound:

- Fine grain size, mostly silt and clay
- 1.1% TOC

Texas, Redfish Bay:

- 99% sand
- 0.1% TOC

Rationale for Toxicity Test Selection

- **Different routes of exposure**

  In pore water:

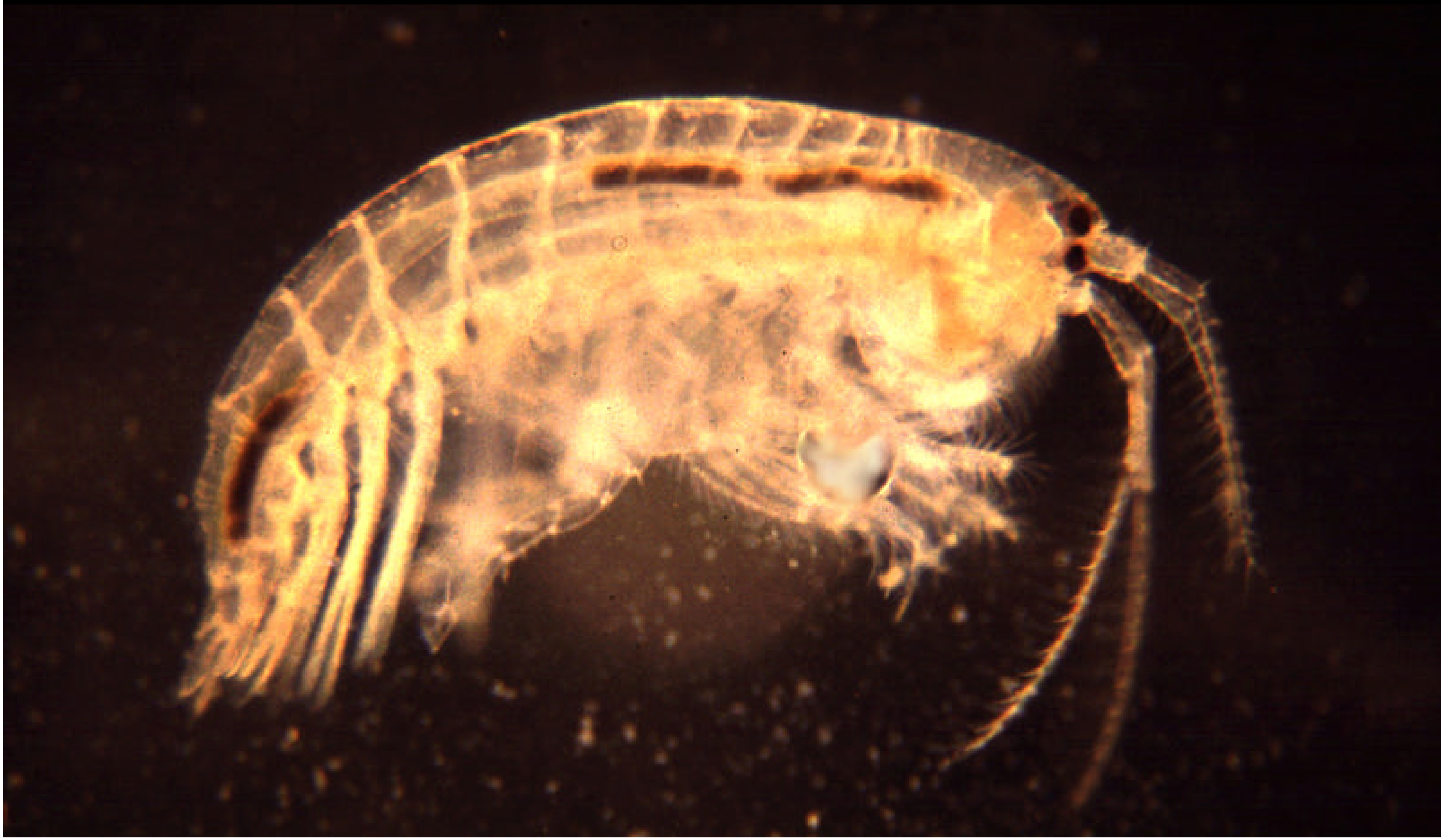
  Not bound to sediment, bioavailable

  Sediment ingestion:

  Bound to sediment, absorbed in digestive tract

- **Most sensitive tests selected based on toxicity database generated in phase I**

Amphipod (*Ampelisca abdita*)



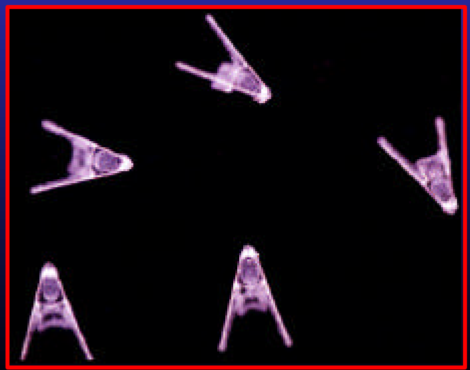
Amphipod test jars: picric acid concentration series


Pneumatic porewater extractors

Most Sensitive Species and Endpoints

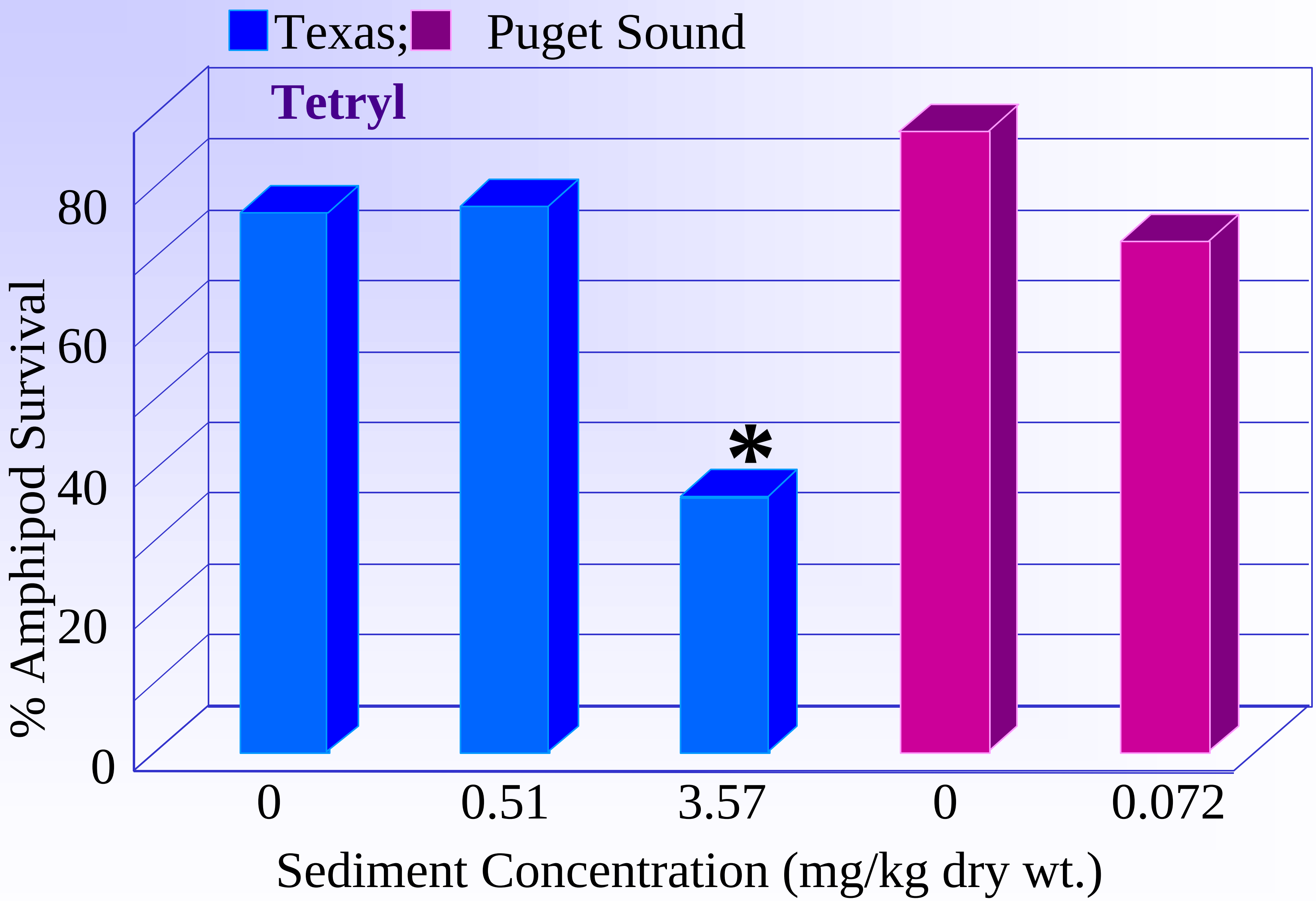
- Polychaete, *Dinophilus gyrociliatus*, reproduction

- Macro-alga, *Ulva fasciata*, germling growth



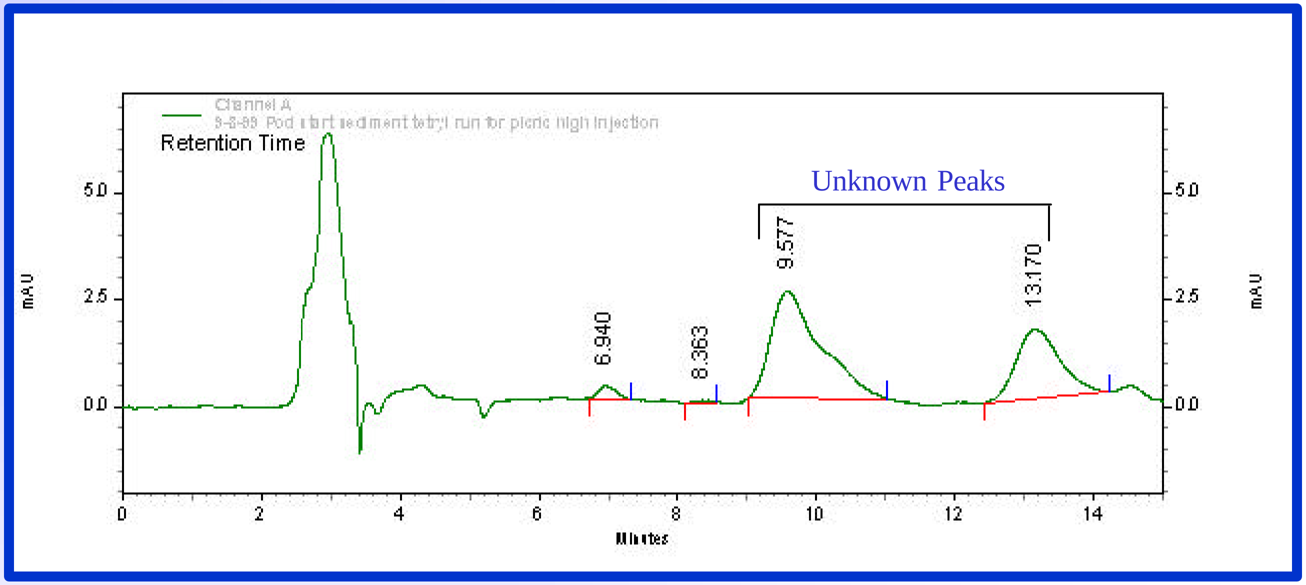
- Sea urchin, *Arbacia punctulata*, embryological development

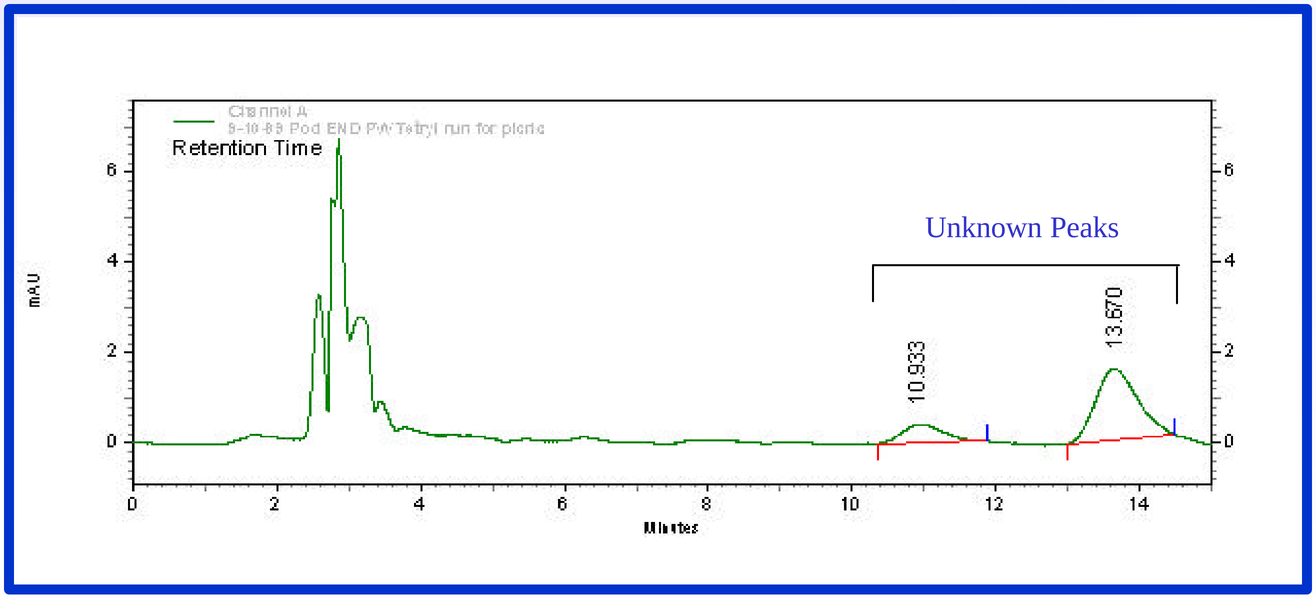
Whole sediment 10-day test with the amphipod, *Ampelisca abdita*



Chromatograms of Puget Sound samples spiked with tetryl, measured using the picric acid method

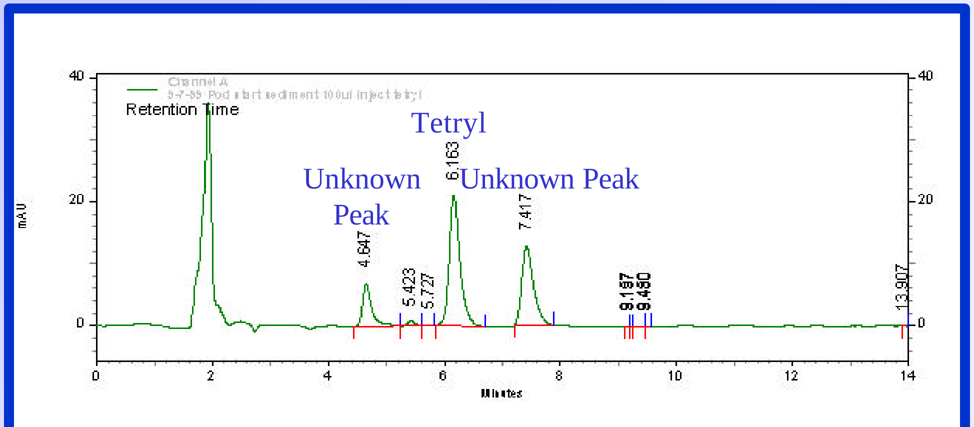
Sediment at amphipod toxicity test start



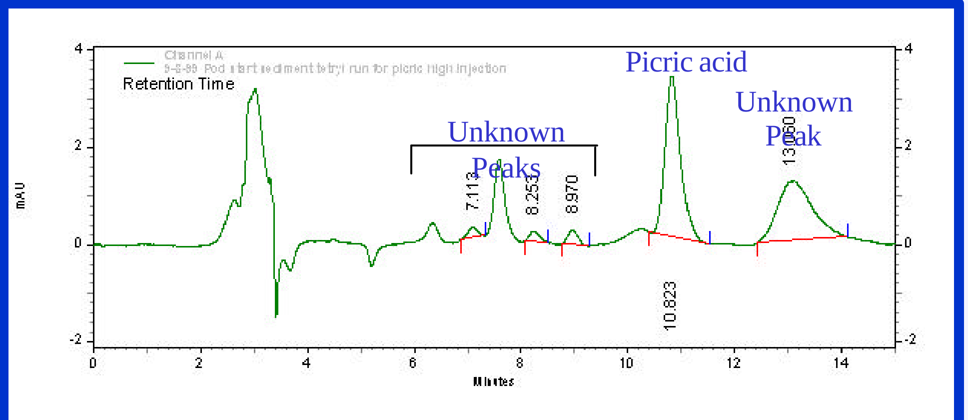


Pore water at amphipod test end

Chromatograms of Texas samples spiked with tetryl

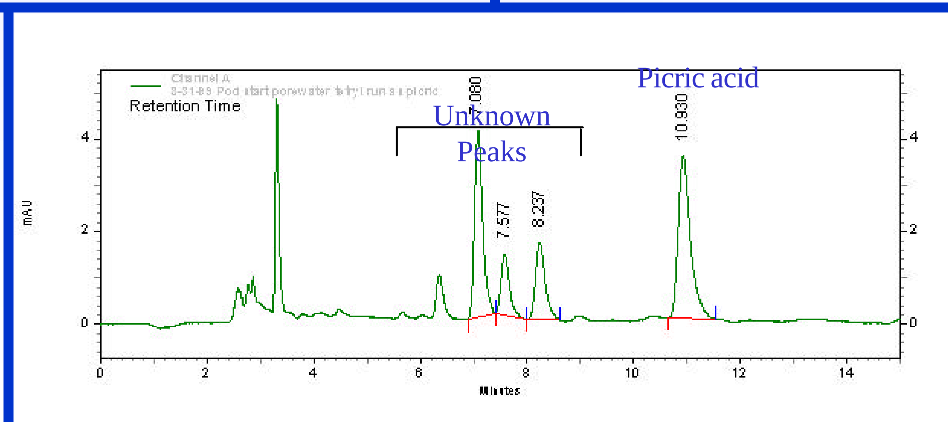


Sediment at amphipod toxicity test start, measured for tetryl

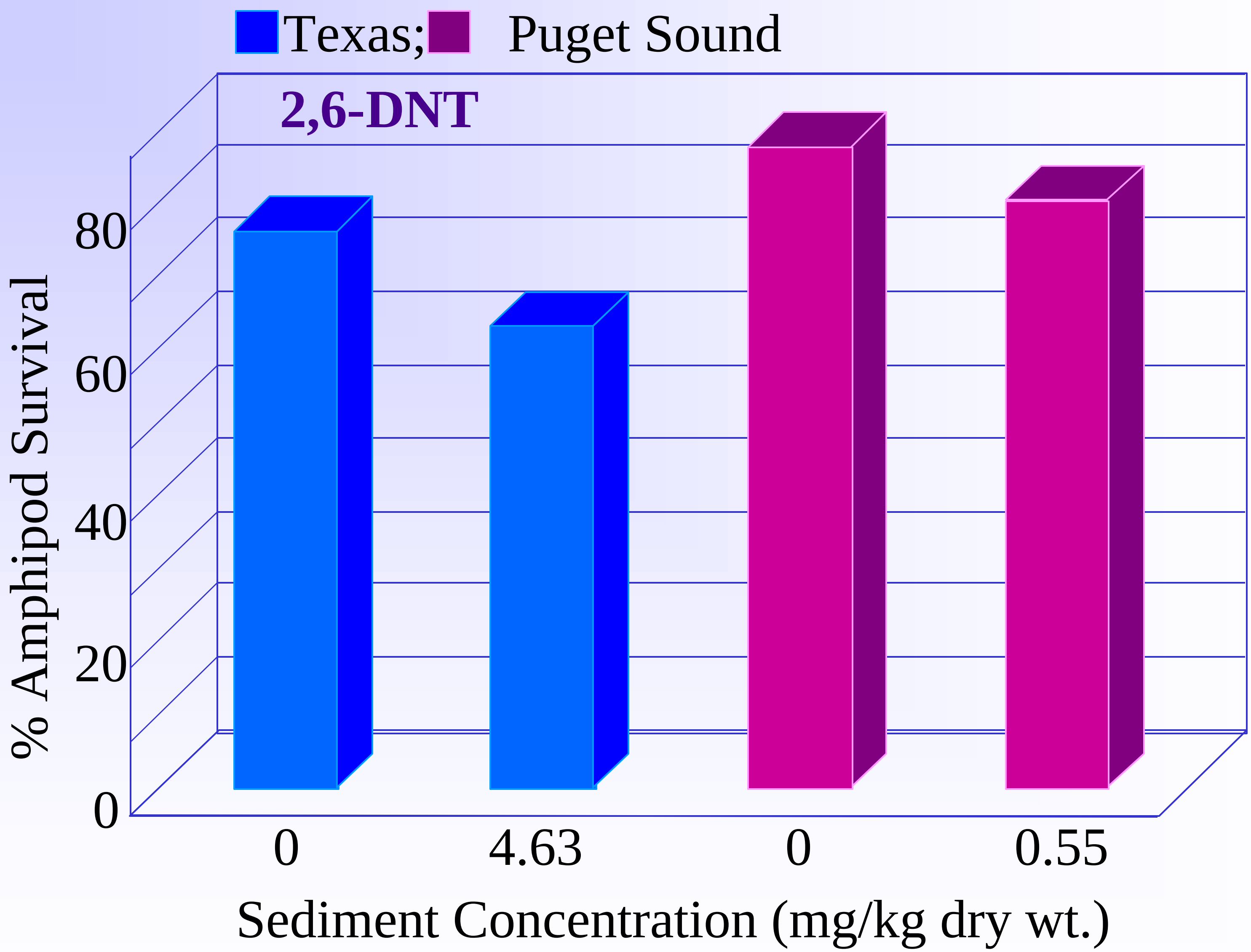


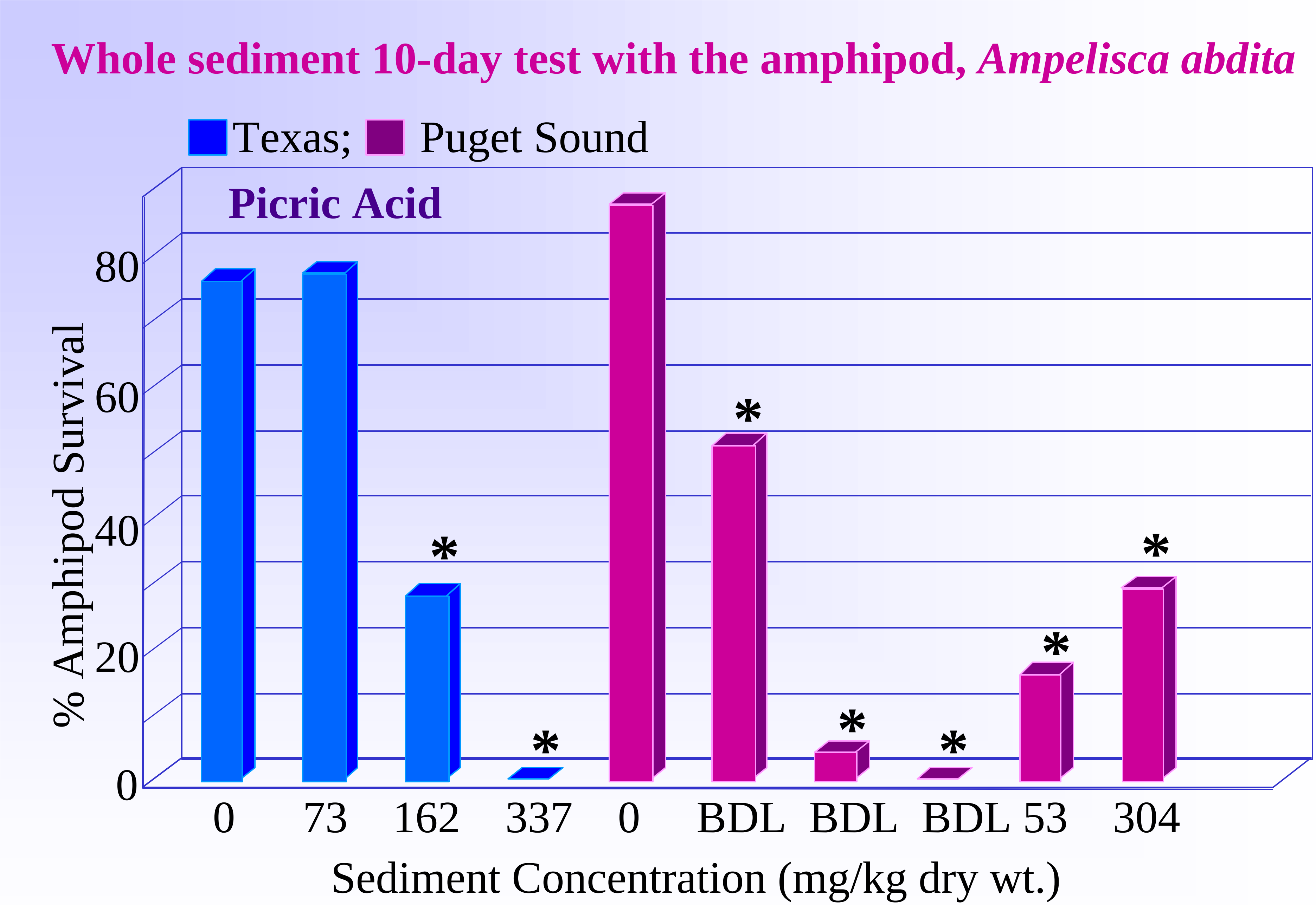
Same as above, but measured using picric acid method

Pore water at amphipod test start, measured for picric acid



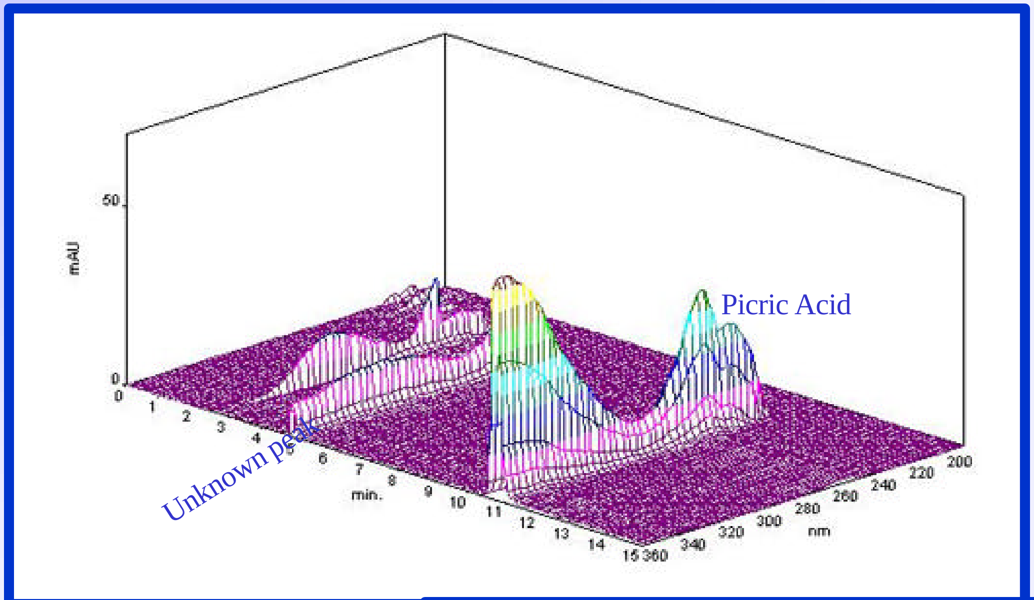
Whole sediment 10-day test with the amphipod, *Ampelisca abdita*

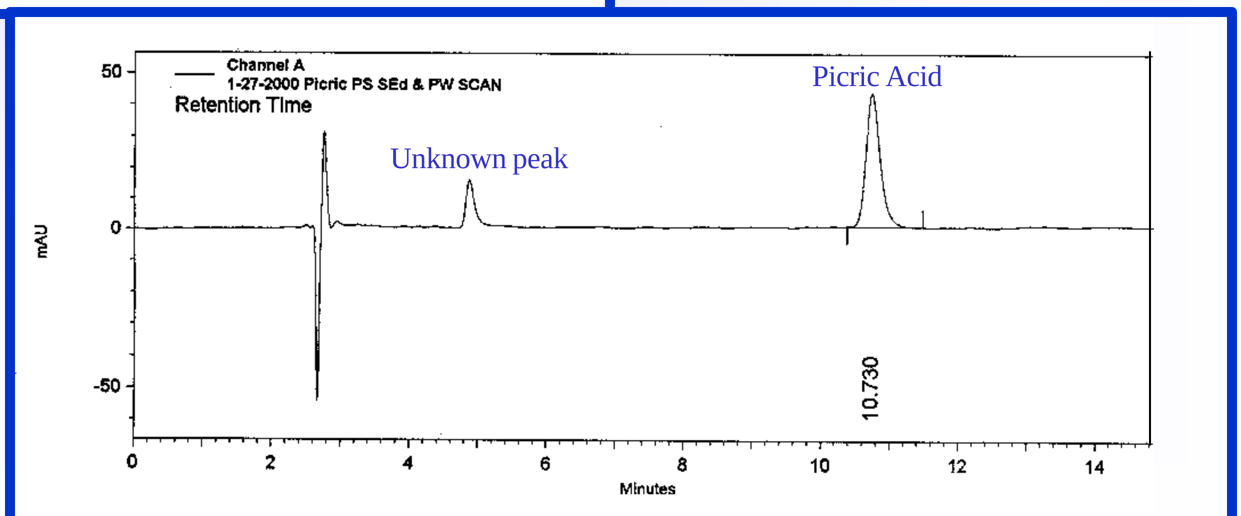




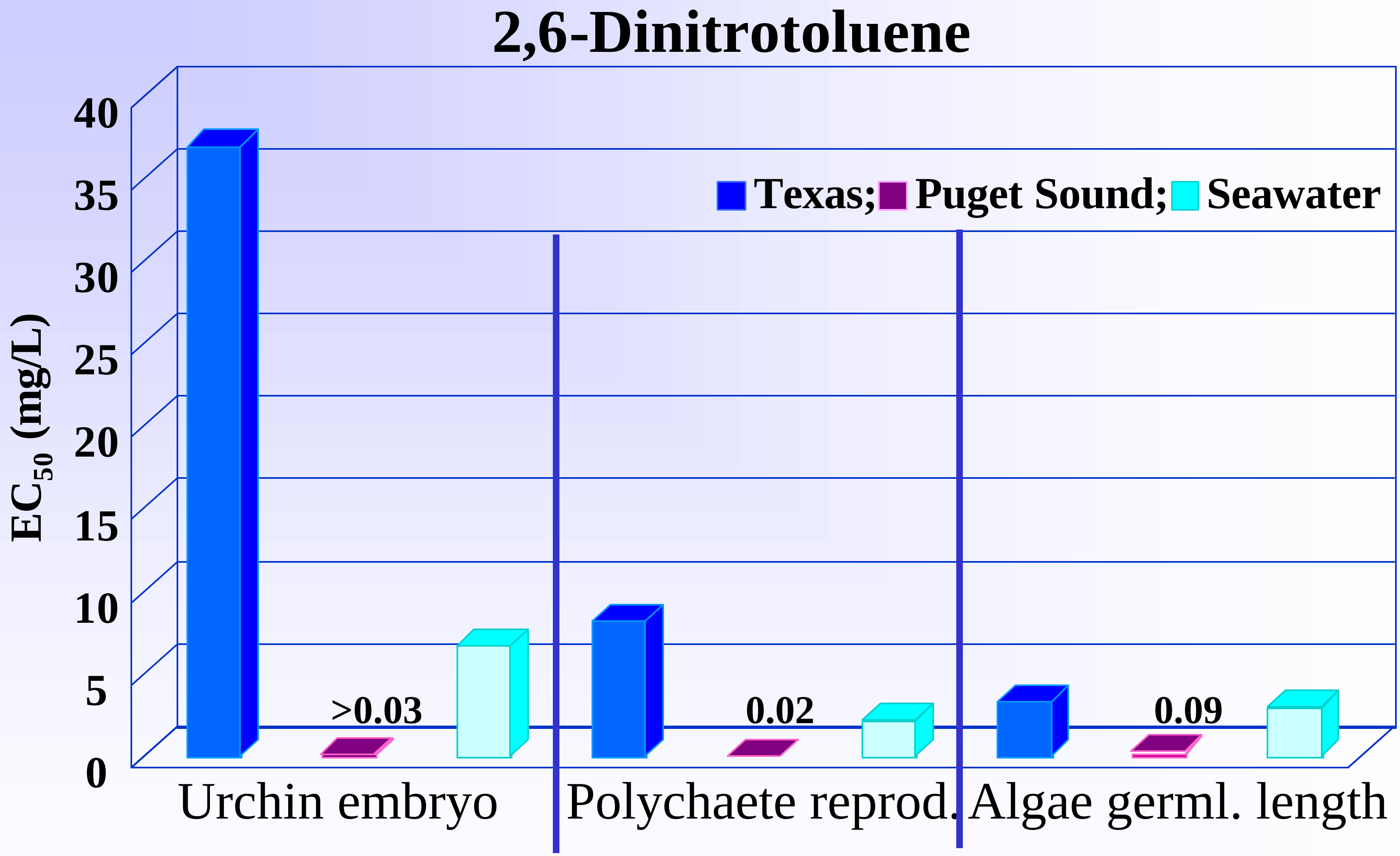
Whole sediment 10-day test with the amphipod, *Ampelisca abdita*
Texas; Puget Sound
Picric Acid
% Amphipod Survival
0
20
40
60
80
*
*
*
*
*
*
*
0
73
162
337
0
BDL
BDL
BDL
53
304
Sediment Concentration (mg/kg dry wt.)

Chromatogram of Puget Sound pore water from sediment spiked with picric acid, showing an unknown peak, possibly a degradation product



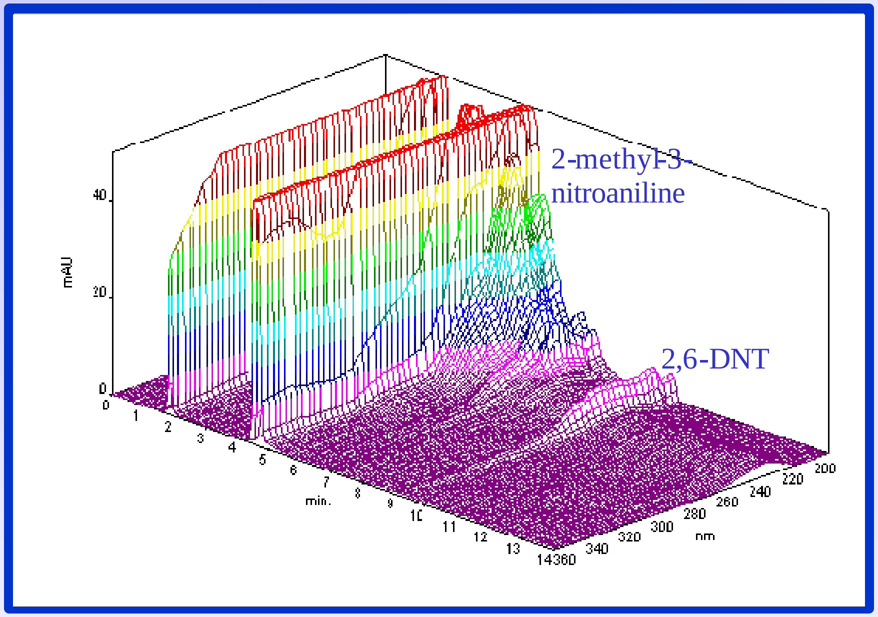


Porewater Toxicity Test Results

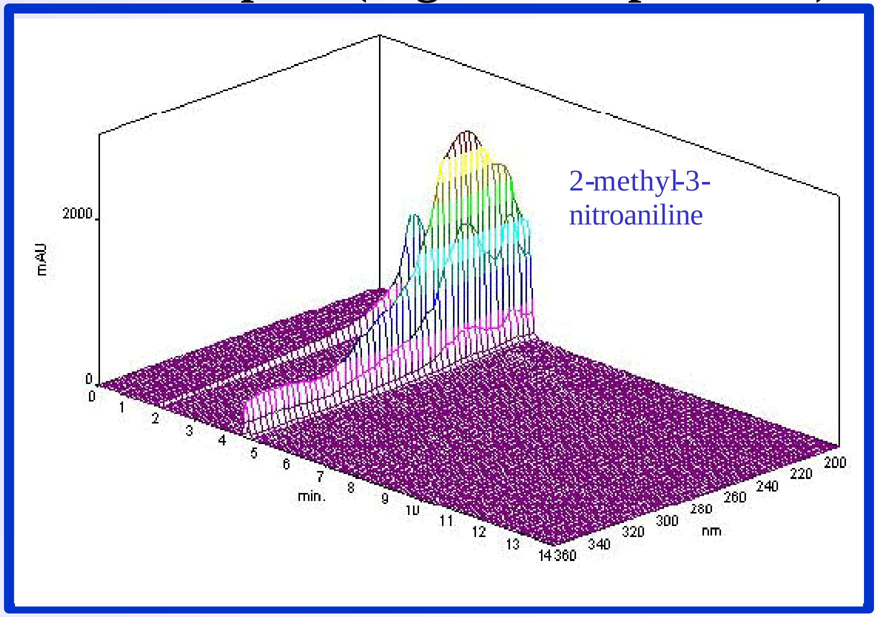


Chromatogram of Puget Sound pore water spiked with 2,6-DNT

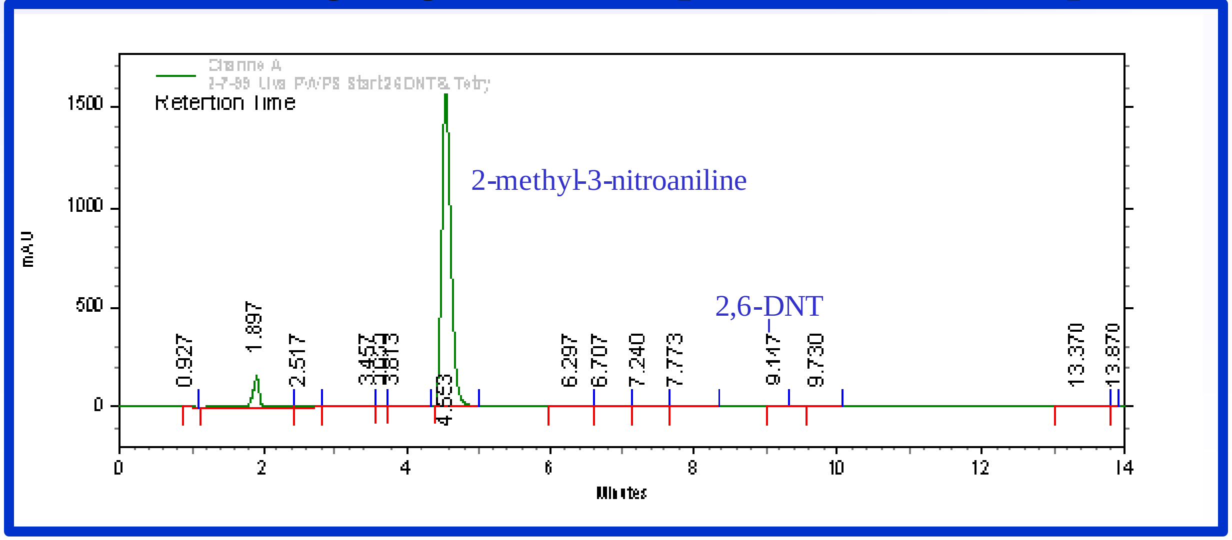
Truncated Y axis for observation of smaller peaks



Expanded Y axis showing entire unknown chemical peak (degradation product?)

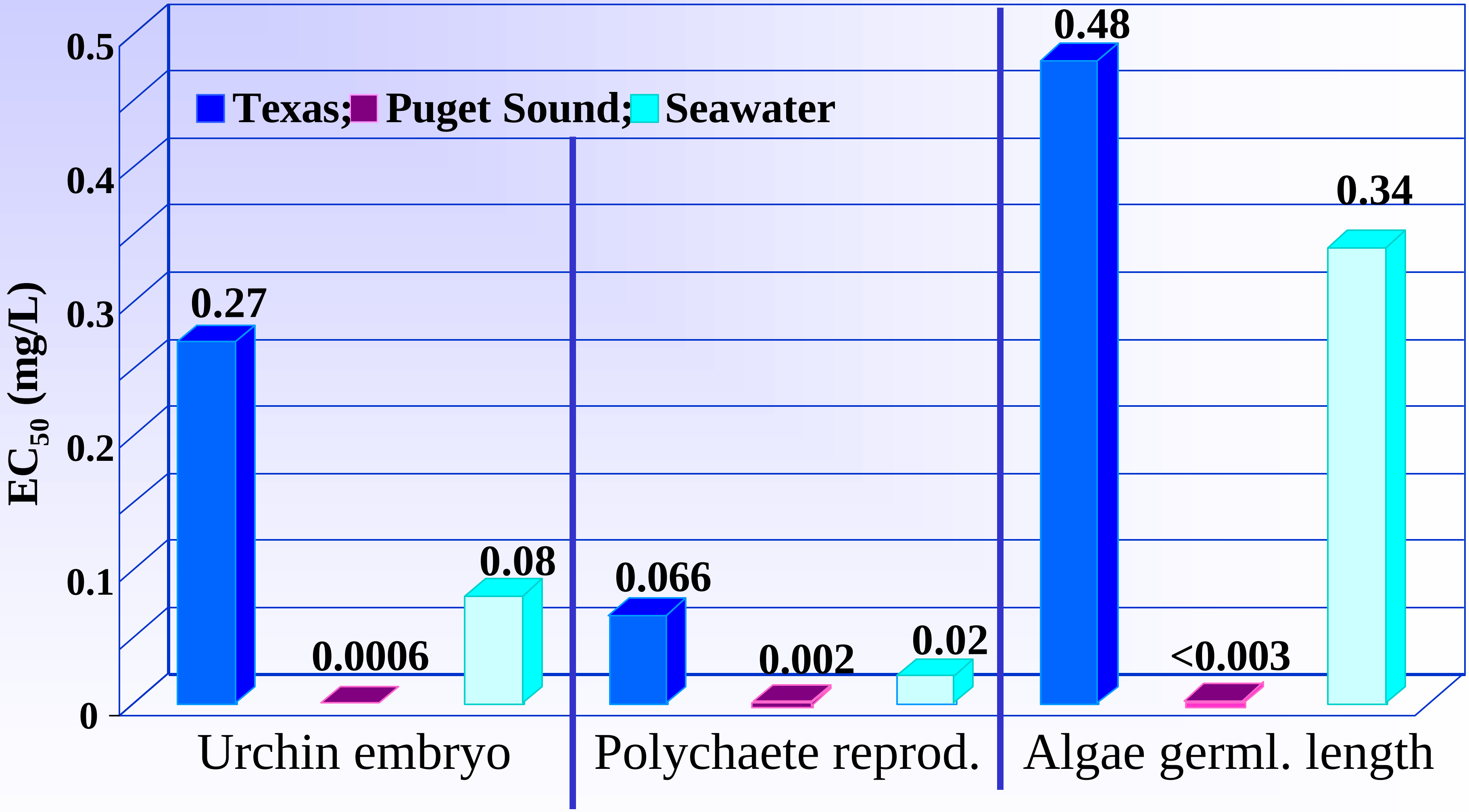


2-D view showing large unknown peak and 2,6-DNT peak barely visible.

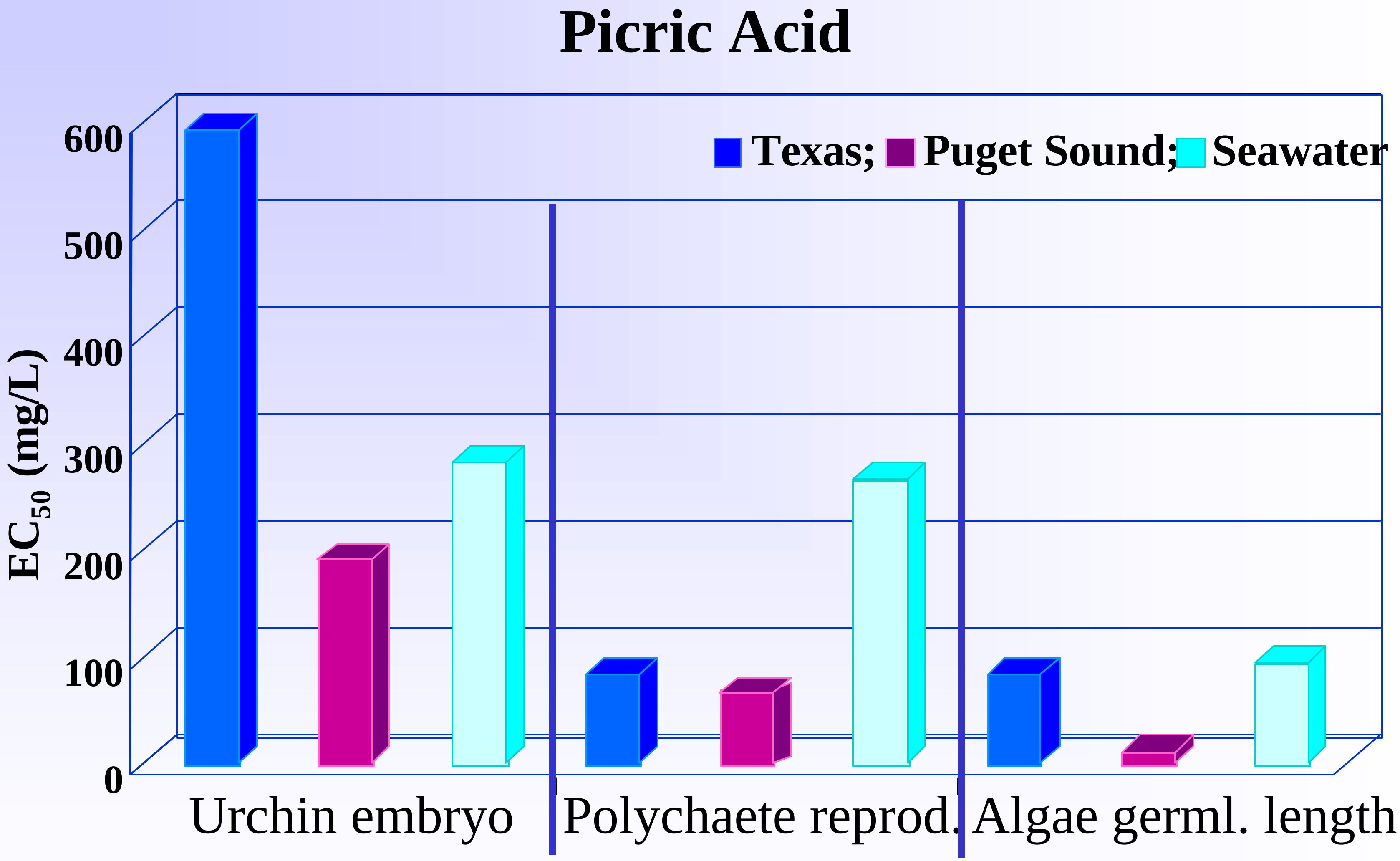


Porewater Toxicity Test Results

Tetryl



Porewater Toxicity Test Results



Summary of Results from Sediment Spiking Studies

- 2,6-DNT, tetryl and picric acid behave differently in different kinds of sediments.
- 2,6-DNT, tetryl and picric acid were either degraded or irreversibly bound in Puget Sound sediment (1.1% TOC).
- Nearly all Puget Sound sediment and porewater samples spiked with ordnance compounds were more toxic than Texas samples or seawater with the same compounds.
- This is likely due to microbial degradation products.
- Nearly all Texas porewater samples were less toxic than filtered seawater spiked with the same ordnance compound, possibly due to sorption by organic carbon.

Overall Conclusion

- It is not sufficient to look at known and expected ordnance compounds in sediments. Degradation products can play a major role in sediment toxicity and effects to the benthic biota.

Recommendations for Future Studies

- Determine the effect of sediment type (grain size distribution and organic carbon content) on the microbial degradation of 2,6-DNT and picric acid (and other ordnance compounds of concern) in marine sediments.
- Evaluate the effect of UV light on the degradation of 2,6-DNT and picric acid (and other ordnance compounds of concern) in marine waters.

Recommendations continued

- Determine if degradation products generated by bio- and photo-degradation of ordnance compounds differ in their nature and are more or less toxic to marine organisms than the parent compounds.
- Assess whether the degradation of ordnance compounds will proceed through complete mineralization in sediments given sufficient time.

Reports

Development of Marine Sediment Toxicity for Ordnance Compounds and Toxicity Identification Evaluation Studies at Select Naval Facilities
http://erb.nfesc.navy.mil/erb_a/restoration/fcs_area/con_sed/tox_marine_sed.pdf

Toxicity of Marine Sediments and Pore Waters Spiked with Ordnance Compounds
http://erb.nfesc.navy.mil/erb_a/restoration/fcs_area/con_sed/marinesed2000.pdf

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